Journal of the Lepidopterists' Society
50(4), 1996, 297–302

NATURAL PUPATION SITES OF THREE NORTH AMERICAN SWALLOWTAIL BUTTERFLIES: *EURYTIDES MARCELLUS* (CRAMER), *PAPILIO CRESPHONTES* CRAMER, AND *P. TROILUS* L. (PAPILIONIDAE)

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ABSTRACT. Pupation sites were determined by releasing wandering prepupal larvae marked with fluorescent paint in a natural area during the day and searching with a UV spotlight the following night. Two of the species studied have environmentally cued pupal color dimorphism: *E. marcellus*, which pupates preferentially on the undersides of leaves, both living and dead, and *P. troilus*, which prefers slender stems among leaves, either living or dead. *Papilio cresphontes* has monomorphic pupae resembling dead wood and prefers pupation sites on dead sticks and woody stems. Together with previous studies, these results support the hypothesis that pupal color determination in swallowtails has evolved in relation to pupation site preference.

Additional key words: pupal color, environmentally cued dimorphism.

Swallowtails, among other groups of butterflies, vary in the ability of their prepupal larvae to respond to environmental cues at their pupation sites. Some species have environmentally cued pupal color polymorphism, with green or brown the most common alternatives, while other species have monomorphic green or brown pupae (Clarke & Sheppard 1972, West et al. 1972, Wiklund 1975, Smith 1978, Hazel & West 1979). The term 'brown' includes shades of gray and really means that the pupa does not resemble a green leaf in its reflectance spectrum. Brown pupae may be variable in color, and in some species may be tinged with dark green (as in *Papilio anchisiades* Fabr., Tyler et al. 1994), but green pupae are usually quite uniform.

We have hypothesized that pupal color determination has evolved in relation to pupation site preference (Hazel & West 1979) and have shown that selection by primarily non-visual predators can drive the evolution of the latter (West & Hazel 1982). The hypothesis predicts that species with sites that are inherently variable in color (for example, leaves or weed stalks that may be alive or dead) will tend to evolve pupal color dimorphism, while monomorphism is expected in species with inherently uniform sites (for example, dead wood in the leaf litter for a brown species, or leafy twigs of evergreen shrubs and trees for a green species).

A major problem testing the hypothesis is the lack of reliable information about pupation sites and pupal colors. We have described pupation sites and pupal colors of *Battus philenor* (L.), *Papilio glaucus* L. and *P. polyxenes* L. (West & Hazel 1979, Hazel & West 1979) and of *P. eurymedon* Lucas (West 1995), from experimental data. Pupal colors have also been described for two other North American species with environmentally cued pupal color, *Eurytides marcellus* (Cramer) (West & Hazel 1985) and *Papilio troilus* L. (Igarashi 1979). We report here experiments on the pupation sites of these two species and of *Papilio cresphontes* Cramer, which has monomorphic pupae (Tyler et al. 1994: 105).

MATERIALS AND METHODS

The insects used in this study were obtained from field-collected larvae and eggs, and from the eggs of field-collected females or those reared and hand-paired in the laboratory. All material originated in Montgomery and Giles counties, Virginia. Larvae were reared in plastic boxes on natural midsummer daylength and temperature and were fed leaves of their local food plants. The experiments were carried out in Blacksburg, Virginia in the summers of 1978 and 1979. The larval food plants of all three species occur locally chiefly in forest understory and along forest edges. They are pawpaw, *Asimina triloba* (L.) Dunal (Annonacaeae), for *E. marcellus*; spicebush, *Lindera benzoin* (L.) Blume, and sassafras, *Sassafras albidum* (Nutt.) Nees (Lauraceae), for *P. troilus*; and hoptree, *Ptelea trifoliata* L., and prickly ash, *Zanthoxylum americanum* Mill. (Rutaceae), for *P. cresphontes*.

To determine the natural pupation sites we used UV-fluorescent paint (West & Hazel 1979). As larvae voided their guts and entered the prepupal stage during the day, they were marked with paint and released in one of three study areas: 'forest,' and two areas of 'edge.' The study areas are described in West & Hazel (1979). In general, the 'forest' area was dominated by trees, with ground cover of dead leaves and sticks and some sparse vegetation. The 'edge' areas were between the 'forest' and an adjacent meadow and had ground cover of tall grasses and weeds as well as saplings. In each area all larvae were released on the same branch of a small tree. Prepupal larvae were located that evening by searching the study areas with a UV spotlight, from the ground to several meters high in the trees. The type of pupation site and its height from the ground were noted, and except for pupations on the undersides of leaves, the diameter of the site was measured at the point of girdle attachment. For dimorphic species, pupal color was recorded after pupation.

Success in finding marked prepupae varied. For *E. marcellus* and *P. troilus* it was about 70%, but for *P. cresphontes* only about 35%. We believe that the low recovery rate, especially of *P. cresphontes*, was due pri-

TABLE 1. Pupation sites of *Papilio cresphontes*, *P. troilus* and *Eurytides marcellus* during the summer in a natural environment. Distributions are different for the three species (χ^2=72.0, df=4, p<0.01).

Species	Pupation site: Dead sticks or living stems, diam.≥0.5 cm	Pupation site: Dead sticks or living stems, diam.<0.5 cm	Pupation site: Leaves
P. cresphontes	11	4	0
P. troilus	0	20	2
E. marcellus	1	3	19

marily to predation on wandering larvae, as fluorescent remains of larvae were often found in the evening on the ground. Once the prepupae had hung up, there was very little loss from predation.

RESULTS

Pupation sites are given in Table 1. *Papilio cresphontes* larvae wandered up to 5 m from the release tree before choosing a site on a broad dead stick or other woody surface above the leaf litter. Eleven of 15 sites were on dead branches more than 0.5 cm in diameter, and the remainder on dead weed stalks and woody stems of the vine *Parthenocissus quinquefolia* (L.) Planchon (Vitaceae). *Papilio cresphontes* has monomorphic brownish pupae resembling lichen-covered twigs, with greenish-grey thoracic and abdominal saddles. *Papilio troilus* larvae moved up to 6 m from the release tree and chose slender stems (20 of 22 sites less than 0.5 cm in diameter) among green or brown leaves. *Eurytides marcellus* larvae moved as far as 3 m, and 19 out of 23 of them chose sites on the undersides of leaves, either living or dead.

DISCUSSION

Taken together with our earlier data (West & Hazel 1979), these results suggest that species with dimorphic pupae (*B. philenor*, *E. marcellus*, *P. polyxenes*, *P. troilus*) use inherently variable pupation sites, but that the variability is in spite of differences among species in the types of sites that they choose. For example (Fig. 1, Table 2), these four species show distinct differences in height above ground, in type of substrate (leaves or stems) or in the width of the substrate, yet in all cases the substrate or its immediate surroundings may or may not be colored by the chlorophylls and xanthophylls of living leaves. The monomorphic species (*P. cresphontes*, *P. glaucus*) also differ somewhat in their sites, but both use exclusively woody substrates.

Among swallowtails, pupal color dimorphism has probably evolved from monomorphic brown (Hazel & West 1995, from physiological evi-

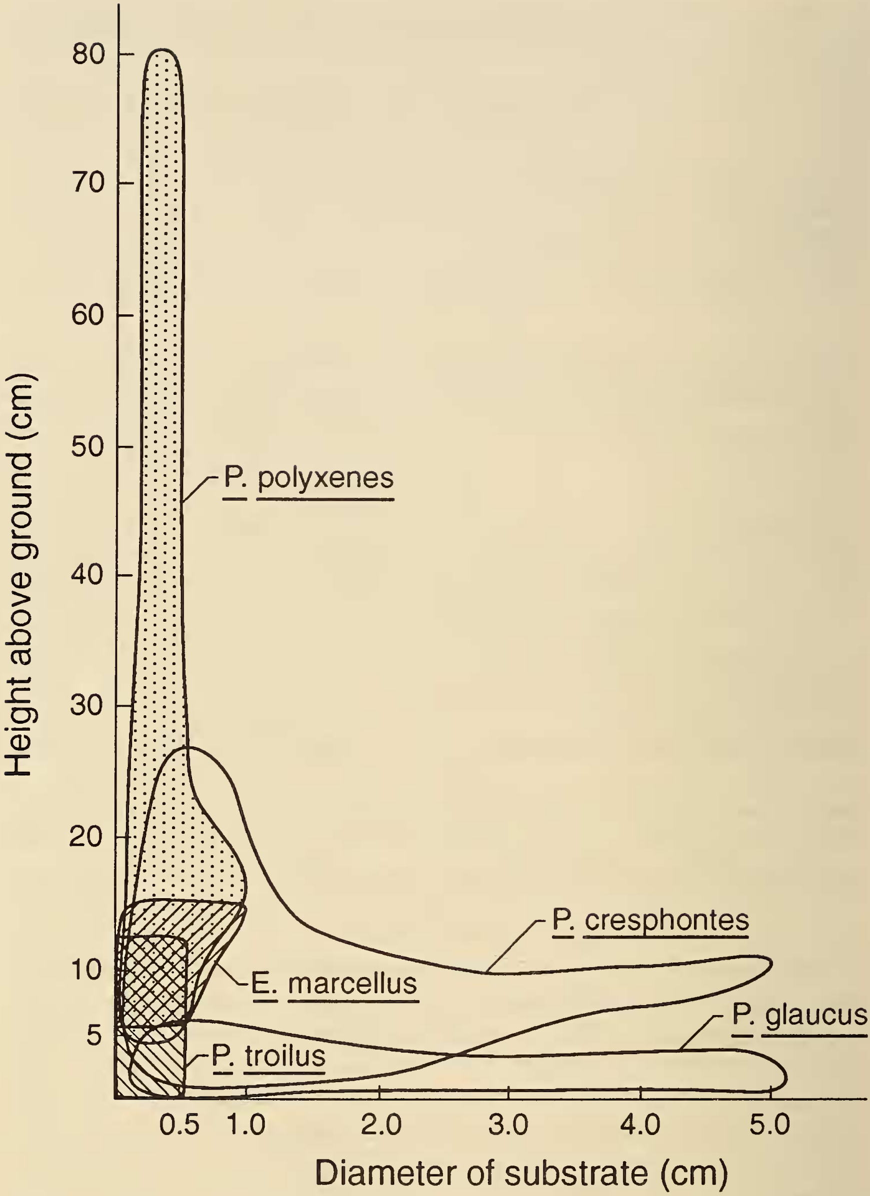

FIG. 1. Ranges in the diameter and height above ground of natural pupation sites of five sympatric swallowtails of eastern North America during the summer generation. The outlines enclose about 95% of the pupations. For pupations on leaves (*Eurytides marcellus* and *Papilio troilus*) only height is given. A sixth species, *Battus philenor*, pupates on vegetation higher than about 90 cm and scarcely overlaps these distributions, but on cliffs may be from 15 cm to several m above the ground or the nearest ledge.

TABLE 2. Variation in pupation sites and pupal color of six eastern North American swallowtails. Data for *Papilio polyxenes*, *P. glaucus* and *Battus philenor* from West & Hazel (1979).

Species	Pupal color	Pupation sites
P. polyxenes	dimorphic	living or dead weed stalks, or woody surfaces above the ground
P. troilus	dimorphic	very slender stems among green or brown leaves close to ground level
B. philenor	dimorphic	tree trunks, cliffs and slender stems well off the ground
E. marcellus	dimorphic	undersides of green and brown leaves close to ground level
P. glaucus	monomorphic brown	undersides of dead sticks or leaves in the leaf litter
P. cresphontes	monomorphic brown	woody surfaces off the ground

dence; but see West 1995, for another view based on genetic evidence). The evidence that it has evolved independently several times is that both monomorphism and dimorphism are found within species groups of the major tribes of swallowtails. In the *glaucus* group of the Papilionini, for example, most species have monomorphic brown pupae, but *P. eurymedon* is dimorphic (West 1995). In the *marcellus* group of the Leptocircini, *E. marcellinus* (Doubleday) has monomorphic brown pupae and pupates in curled-up dead leaves (Turner, *in* Collins & Morris 1985), whereas *E. marcellus* is dimorphic. In the Troidini, some *Battus* species are dimorphic, for example *B. belus* (Cramer) and *B. crassus* (Cramer) (Tyler et al. 1994), and others are monomorphic green (e.g., *B. lycidas* (Cramer)) (Moss 1919). Among Old World troidines, at least some *Troides* spp. are dimorphic, but all members of *Ornithoptera* appear to be momomorphic brown (Igarashi 1979).

ACKNOWLEDGMENTS

This research was supported by National Science Foundation grant DEB 78-02474 to D. A. West, and the Department of Biology, Virginia Polytechnic Institute and State University.

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Received for publication 15 April 1995; revised and accepted 5 January 1996.